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Molecular basis of colorectal cancer – role of gastrin and cyclooxygenase-2

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key words: gastrin, gastrin receptors, COX-1, COX-2, Helicobacter pylori, apoptosis, colorectal cancer

SUMMARY

Background: *Tumors arising in the colorectal area have worldwide distribution and concern mostly older population being attributed to genetic, dietary and hormonal factors but most recently also to infection with Helicobacter pylori (HP). Both, HP discovery and molecular biology of colorectal cancer have been recently considered as two of ten greatest advances of gastroenterology at the dawn of 3rd millenium but little information is available regarding the relationship between the HP and colorectal cancer. Since HP infection is usually accompanied by an increase in plasma level of gastrin, which is also recognized as a trophic hormone for the colonic epithelium and a potent mitogen capable to induce cyclooxygenase-2 (COX-2), we decided 1) to compare the seroprevalence of HP, its cytotoxic protein, CagA, and cytokines (TNFα, IL-1β and IL-8) in colorectal cancer patients, before and after removal of cancer, with those in age- and gender-matched controls; 2) to determine the gene expression of gastrin and gastrin receptors (CCK_B-R) in colorectal cancer tissue, 3) to assess the plasma levels and tumor tissue contents of gastrin, 4) to examine the mRNA expression of cyclooxygenase COX-1 and COX-2 cancer tissue and intact colonic mucosa.*

Material and Methods: *The trial material included 80 patients with colorectal cancers and 160 age- and gender-matched controls. Anti-HP IgG, anti-CagA IgG seroprevalence and cytokine levels were estimated by ELISA tests. Gene expressions of gastrin, CCK_B-R, COX-1, COX-2 and Bax and Bcl_2 was examined using RT-PCR, while gastrin was measured by RIA.*

Results: *The HP IgG seroprevalence, especially that expressing CagA, was significantly higher in colorectal cancer patients than in controls and did not change one week after tumor resection while plasma cytokines were significantly reduced after this operation. Gastrin and CCK_B-R mRNA were detected in the cancer tissue and the resection margin and similarly COX-2 mRNA was expressed in most of cancers and their resection margin but not in intact colonic mucosa where only COX-1 was detected. The colorectal cancer tissue contained several folds more immunoreactive gastrin than cancer resection margin and many folds more than the intact colonic mucosa.*

Conclusions: *1) Colorectal carcinoma and its resection margin overexpress gastrin and receptors for gastrin (CCK_B-R), and COX-2; 2) here, we propose that an increased plasma level of gastrin should be considered as suitable biomarker of colorectal cancer, 3) HP infection may contribute to colonic cancerogenesis by enhancing expression of gastrin and COX-2, they may account for stimulation of the tumor growth, angiogenesis and reduction in apoptosis as evidenced an increased ratio of mRNA expression for anti-apoptotic Bcl_2 over proapoptotic Bax proteins and 4) HP positive patients who develop colorectal cancer should be subjected to the HP eradication; this is expected to reduce hypergastrinemia and to attenuate COX-2 expression. Our final conclusion would be: treatment of patients with colorectal cancer with COX-2 selective inhibitors now gained a strong support as a preventive measure.*

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BACKGROUND

Numerous epidemiological studies demonstrated that malignant tumors in the colorectal area are the most common visceral malignancies concerning mainly old adults with the highest death rates in United States, Australia and Europe including Poland [1,2]. In Poland, over 100, 000 new malignancies are diagnosed with an yearly increase by 2–3% and with colorectal tumors representing about 6–7% of all malignant tumors (2) (Fig. 1). Statistical evidence established a positive relationship between the occurrence of colorectal carcinoma (CC) and dietary factors, especially excessive energy intake, low content of unabsorbable vegetable fibers, intake of red meat, decreased intake of protective microelements and a close association of this cancer with colonic adenomas [3,4]. Besides geography and diet, an age, a family history of CC disease, the presence of chronic colitis and adenoma in distal colon and rectum increase the risk of CC.

The sequence of pathological change from normal mucosa to cancer is schematically presented at Fig. 2. As in other cancers, various genetic alterations have been detected by the time the colorectal tumor becomes clinically apparent including the loss of methyl groups in DNA (hypomethylation), occurrence of activated oncogenes such as K-ras, allelic loss on 18q21, termed DCC (deleted in colon cancer), mutation and inactivation of p53 gene playing the critical role in colon cancerogenesis [5]. Furthermore, an overexpression of COX-2 was reported in colorectal cancer by numerous investigators suggesting that this enzyme could be responsible for abundant release of prostaglandins (PG), angiogenesis and reduction in apoptosis [6–15] similarly as in gastric cancer [16,17] (Fig. 3).

It is well known that the colon embryologically arises from the same endoderm cells that form the lining of the stomach and possesses similar neuro-endocrine and paracrine cells releasing various hormonal peptides and their receptors including gastrin, that has potent mucosal growth promoting effect on colon in animals [18] and humans [19].

Helicobacter pylori (HP) is known to be accompanied by excessive and prolonged release of gastrin that has been suggested to account for the development of gastric cancer [20–22] and that has been shown to coexpress with COX-2 [22]. Since HP infection was found to be accompanied by an overexpression of immunoreactive gastrin that has proliferative influence on colonic mucosa [18,19] we decided to assess the relationship between HP infection and related gastrin release and COX-2 expression in colorectal cancerogenesis. Furthermore, CC cells have been shown to express gastrin [23], therefore, the determination of gastrin in plasma and CC tissue was projected to be determined in our CC patients. As gastrin is a potent mitogen (23) that could induced upregulation of COX-2 and increase the release of PG that in turn might stimulate angiogenesis and tumor growth [8–15], it was rationale to determine the expression of both gastrin and COX-2 in the colorectal carcinoma.

The aims of this study were following; 1) to compare the seroprevalence of HP and its cytotoxic protein, CagA, in colorectal cancer patients with that in age- and gender-matched healthy controls;

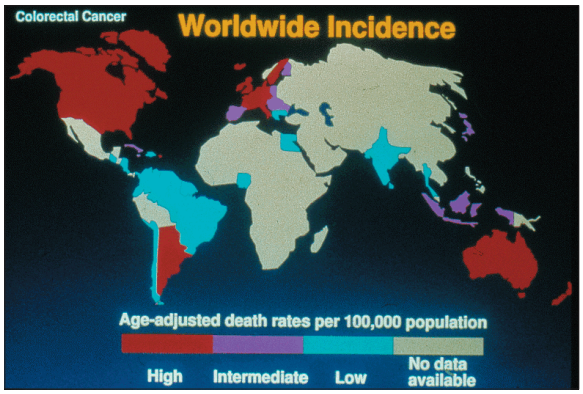

Figure 1. Colorectal cancer is more common in industrialized 'Western' countries, including Poland, with diets that are high in meat, animal fat and total calories but poor in unabsorbable vegetable fibers.

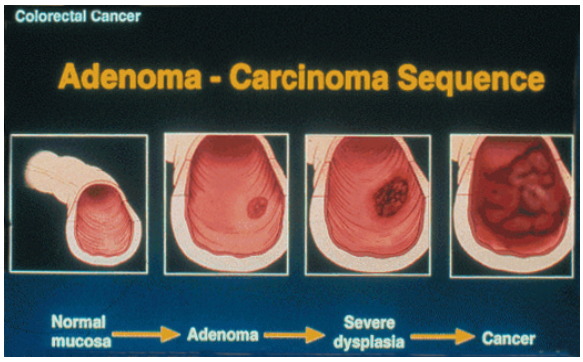

Figure 2. Schematic presentation of the sequence of pathological changes from normal mucosa, through adenoma, 'carcinoma in situ' or severe dysplasia to colorectal cancer though the latter rarely may arise de novo without preceding adenoma

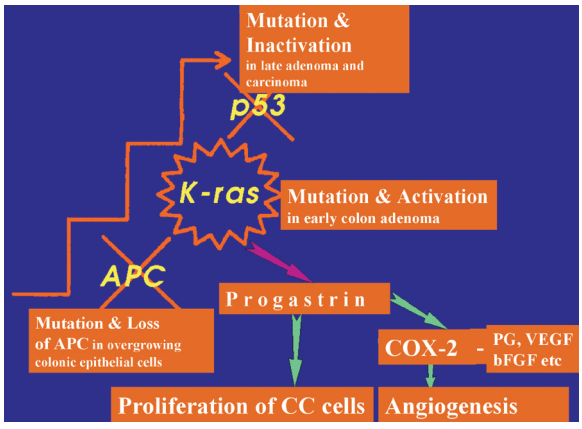

Figure 3. Major steps in cascade of colorectal cancerogensis

2) to determine the gene expression of gastrin and gastrin receptors (CCK_B-R) in colorectal cancer, its resection margin and, for comparison, in antral mucosa; 3) to assess the gastrin content in the plasma of cancer patients and controls as well as in the colorectal tissue, its resection margin and intact colonic mucosa and 4) to examine the mRNA expression of COX-1 and COX-2 as well as the proapoptotic Bax and antiapoptotic Bcl_2 protein expression in the colorectal cancer and its resection margin.

We found that the colorectal cancer and its resection margin contains higher amounts of gastrin than the intact colonic mucosa and that cancer and its resection margin are capable of expressing mRNA for gastrin and gastrin-receptors (CCK_B-R), for COX-1 and COX-2 and for anti-apoptotic Bcl_2 rather than proapoptotic Bax and to release large amounts of immunoreactive gastrin that could locally stimulate (by autocrine or paracrine pathway) the tumor cell proliferation and possibly also to induce COX-2 expression in the cancer and surrounding mucosa.

MATERIAL AND METHODS

The studies were carried out on 40 histologically verified colorectal carcinoma (35 men and 5 women) with the median age of 64 years (range 47 to 82 years) and 160 controls (130 men, 30 women) with the median age of 63 years (range from 47 to 82 years). Patients symptoms, age, gender, tumor site and its histology were recorded at

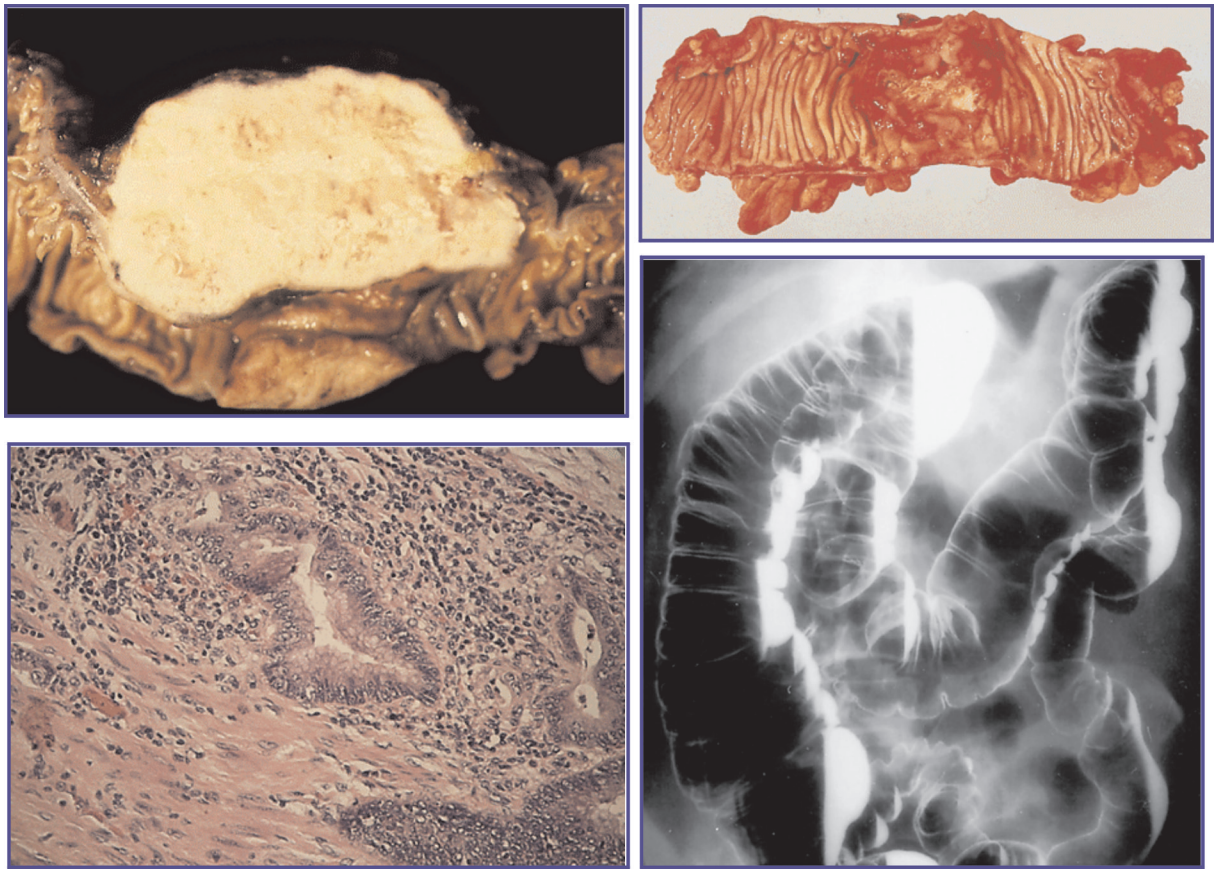

Figure 4. Examples of colorectal cancers of ascending colon and sigmoid macroscopical and microscopical or radiological examinations.

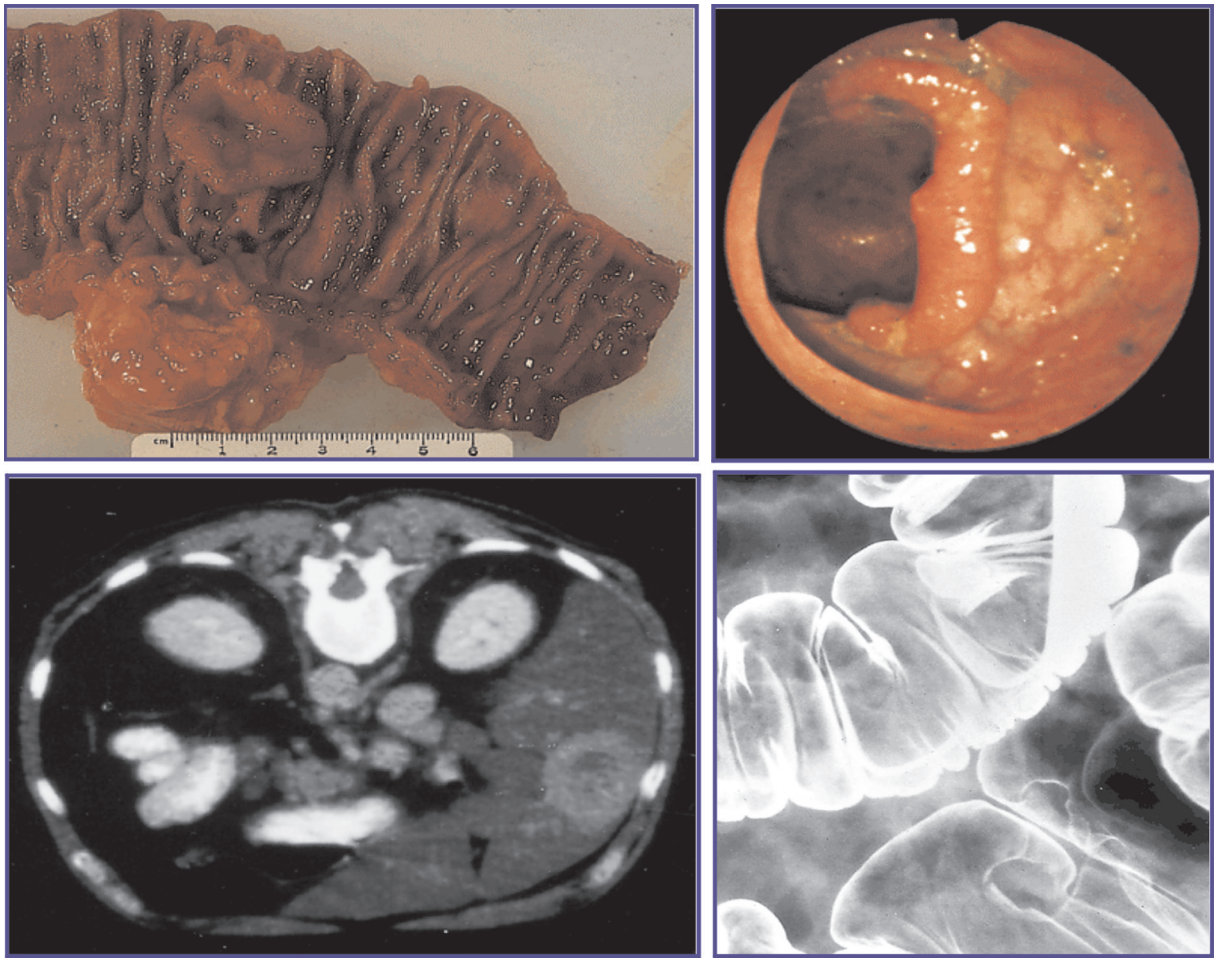

Figure 5. Examples of colorectal cancer of ascending sigmoid macroscopically and in CT (on left) and of caecum at endoscopy and radiology (on right)

the Department of General Surgery of District Hospital of Cracow, Department of Gastroenterology of Pomeranian Medical University, Szczecin and Department of Medicine, University of Erlangen-Nuremberg, Erlangen, Germany and Department of Physiology, Jagiellonian University School of Medicine, Cracow, Poland. Before the surgery, gastroscopy was performed to obtain mucosal biopsy from the gastric corpus and the antrum for histology and for comparison of the mRNA expression of gastrin and its receptors (CCK_B-R) with those in colorectal cancers. During the surgery, the large biopsy samples were taken from the tumor, the resection margin and intact colonic mucosa at the remote site from the cancer for routine histological examination to determine the stage and histological type of the tumor and for gene expression of gastrin and its CCK_B-R, COX-1 and COX-2 and Bax and Bcl_2. The tumors were identified histologically as colorectal tubular (70%) and tubulovillous carcinoma (30%) [5]. Examples of macroscopic, endoscopic, CT and microscopic pictures of colorectal cancers are presented on Figs 4 and 5.

The HP infection status was assessed by our modification of capsulated minidose ^{13}C-Urea Breath Test (UBT) as described earlier [24] and/or by determination of IgG antibodies to HP by enzyme linked immunosorbent assay (ELISA) using commercially available kit (EIAGEN HP IgG, Clone Systems, Italy). Titers higher than 15 AU/ml were considered positive (following the manufacturer recommendations). IgG antibodies against CagA were detected by ELISA using recombinant CagA kindly provided Ora Vax Cambridge, USA as previously [21,22,24].

For the determination of the mRNA expression of gastrin, CCKB-R, COX-1 and COX-2 and Bax and Bcl_2 the analysis of reverse transcription-polymerase chain reaction (RT-PCR) was applied and the tissue content of gastrin was measured using

RIA. For this purpose larger tumor (~100 mg), tumor resection margin (~100 mg) and intact colonic mucosa samples were taken and frozen immediately in the liquid nitrogen for the detection of the signals for gastrin and gastrin receptors as well as COX-1 and COX-2 and Bax and Bcl_2 by reverse transcriptase-polymerase chain reaction (RT-PCR) as described earlier [22,25,26]. Tumor, resection margin and intact mucosa samples were also homogenized in phosphate buffer at pH 7.7 and 0°C for 10 s with Ultra-Turrax T-25, Ika Labortechnik, Staufen, Germany. The homogenized samples were processed as for plasma gastrin RIA using gastrin antiserum, 4562 (kindly donated by Professor J.E. Rehfeld of Copenhagen, Denmark) in the final dilution of 1:500 000. The antibody 4562 recognize amidated G-13, G-17 and G-34 equally. The sensitivity of the present assay was 2.5 pmol/mL plasma equivalent to human G-17. Blood samples were collected preoperatively and in some patients also one week after the surgery from peripheral vein under basal conditions and after separation of the plasma they were stored at -70°C for gastrin RIA as described before [21,27].

RT-PCR and detection of amplified gene products were performed using the above mentioned large biopsy specimens obtained during surgery. For this purpose, total cellular RNA was isolated from these specimens using TRIzol Reagent (Gibco BRL, UK). Briefly, tissue samples placed in to the liquid nitrogen and then transferred to TRIzol Reagent and homogenized. After addition of chloroform the aqueous phase was separated by 13000 rpm spin in 4°C for 10 min. RNA was precipitated using isopropyl alcohol, washed with 75% ethanol and resuspended in DEPC treated water. 1 mg of total cellular RNA was used for the synthesis of cDNA in RT-PCR reaction (Promega Reverse Transcription System). The final concentration of components in 20 ml of total reaction volume was as follows; 5 mM $MgCl_2$, 1x Reverse Transcription Buffer (10 mM Tries-HCl, 50 mM KCl, 0.1% Triton X-100), 1 ml each dNTP 1 U/ml Recombinant RNasin Ribonuclease Inhibitor, 15 U/mg AMV Reverse Transcriptase 0.5mg $Oligo(dT)_{15}$ Primer per microgram RNA. Samples were incubated at 42°C for 15 minutes and heated at 99°C for 5 minutes followed by a 5 min incubation at 0–4°C.

The PCR reactions were prepared in 1 x Dynazyme PCR buffer (Flowgen) with 40 mmol dNTPs (Pharmacia) and 10 mmol of upper and lower primers. The following primer pairs for PCR reaction were used: 5′-GCC CAG CCT CTC ATC ATC as a sense primer 3′-GGG GAC AGG GCT GAA GTG - antisense for gastrin; 5′-TCT CGC GAG CTC TAC TTA GGG as sense primer and 3′-GAA GTT GCA CGT AGC AGC CA - antisense for CCK_B-R; 5′-AGC GGG AAA TCG TGC GTG as sense primer and 3′-GGG TAC ATG GTG GTG CCG as antisense for β-actin; 5′-GCA ACA CTG GAA CAT GGC TA-3′ as a sense primer and 5′ACG CCA CCA TTC TGT CTT TG-3′ as an antisense primer for COX-1; and 5′-TCA TTC ACC AGG CAA ATT GCT GGC AGG G-3′ as a sense primer and 5′-ACA GTT CAG TCG AAC GTT CTT TTA GTA GTAC-3′ as an antisense primer for COX-2, 5′ TGG CAG CTG ACA TGF TTT CTG AC as sense primer and 3′ CGT CCC AAC CAG GGT CT as antisense for Bax, 5′ CAG ATG CAC CTG ACG CCC TT as sense primer and 3′ CCC AGC CTG CGT TAT CCT GGA as antisense for Bcl_2, as described before [26].

The thermal profile for all these reactions was the same; 95°C for 5 min before 40 cycles or 95°C for 45 s, 60°C for 90 s, and 72°C for 90 s. This was followed by a single stage of 60°C for 120 s and 72°C for 180 s. PCR products were analyzed by agarose gel electrophoresis and abundance of cDNA in each sample was estimated by video densitometry analysis (Fotodyne, USA) using Gel-Pro Analyzer program and expressed as a ratio versus calculated intensity of β-actin product used as a reference gene. In the case of the gastrin gene products the intensity of two bands (215 bp and 345bp) were summrized and the same procedure was applied to express the results.

Statistical analysis

The analyzed groups were those with HP-, CagA-seropositivity and plasma levels and tissue immunoreactive gastrin contents in patients with gastric cancer and controls. In general, rank sum test, Sperman's rank test order correlation was used for relation between independent variables. A P value of less than 0.05 was accepted as significant.

RESULTS

The prevalence of HP seropositivity reached ~85% in colorectal cancer patients but only 60% in controls and this difference in HP seropositivity between cancers and controls was statistically significant. The CagA seropositivity in colorectal cancer patients was about twice as high (42%) as in controls (20%) and this difference was also statisti-

cally significant. The UBT in cancer patients was positive only in 65%, while in controls it was positive in 57% of tested cases.

Basal plasma levels of TNFα, IL-1β and IL-8 showed significant decrease one week after the removal of colorectal cancer but the HP and CagA seropositivity remained the same as before the surgery and these results are not included.

Basal plasma amidated gastrin levels in colorectal cancer patients were about 53±12 pM and they were about over twice as high as those in controls (21±5 pM). One week after removal of colorectal cancer, the plasma gastrin level was reduced by about 20% significantly altered (40±6 pM before surgery vs 25±3 pM after surgery). The gastrin contents in colorectal cancer tissue were about 6.5 and 5 pmol/g of wet tissue weight and it was about three times higher than at the resection margin and 20 times more than in the macroscopically normal antral mucosa. This difference between the plasma gastrin concentration in cancer patients and in healthy controls as well as between the gastrin content in tumor tissue, resection margin and intact colonic mucosa was highly statistically significant. Fig. 6 shows the diagram illustrating the posttranslational processing of pre-progastrin into progastrin

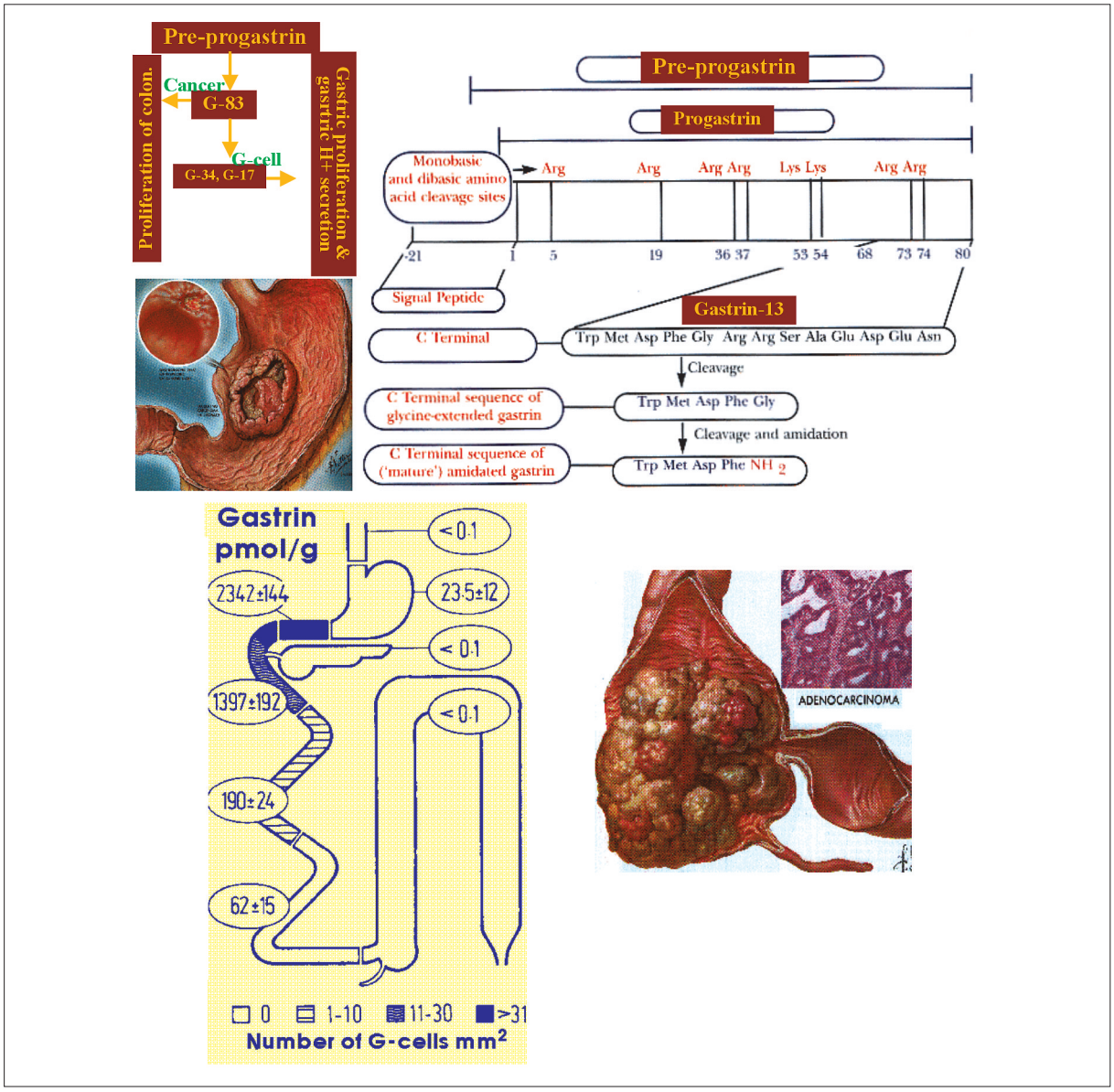

Figure 6. The post-translational processing of pre-progastrin into progastrin and amidated gastrins (G-13, G-17, G-34) in G-cells located mainly in antrum and duodenum and into progastrin in colorectal cancer (on right)

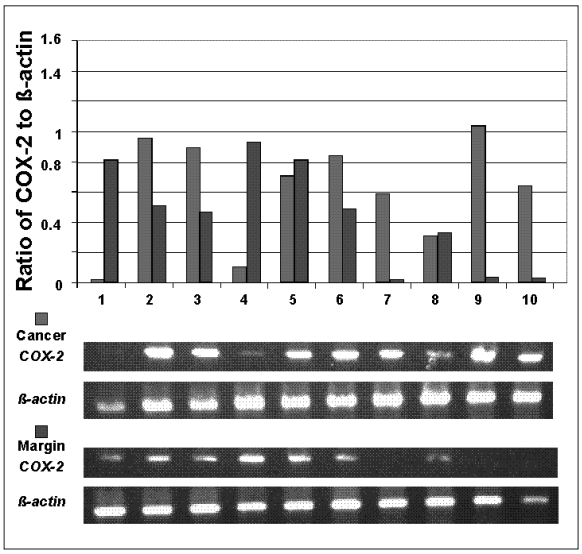

Figure 7. Expression of COX-2 and actin and ratio of COX-2 to actin in colorectal cancer and its margin

in colorectal cancer cells and in amidated gastrins (G-13, G-17 and G-34) in G-cells in antro-duodenal area and in gastric cancer cells.

The tumor tissue of all cancer patients showed mRNA expression for gastrin, though this expression was relatively small in 3 out of 12 examined cancers, and similar expression was detected in antral biopsy samples taken from these patients. The ratio of gastrin mRNA to β-actin mRNA varied from case to case and it was in 8 of 12 examined cases somewhat higher in the cancer tissue than in resection margin and similar to that in the intact antral mucosa. The mRNA expression for gastrin receptor (CCK_B-R) signal was observed in all except one tumor and in 8 of 12 resection margins of these tumors and in case of cancer tissue it was similar to that in intact mucosa of gastric corpus. The ratio of CCK_B-R mRNA to β-actin mRNA was higher in the majority of cases in tumor tissue than in resection margin and reached the values similar to that in intact mucosa of gastric corpus of these patients. This ratio was negligible in the antrum mucosa and these results have been omitted for the sake of clarity.

The COX-1 mRNA was expressed in the cancer tissue and the resection margin as well as in intact colonic mucosa. COX-2 mRNA expression was pronounced in all, except one, of the cancer tissue samples and in 9 of 12 of the resection margins but was not detectable in the intact colonic mucosa. In 8 of 12 cases tested, the COX-2 mRNA expression was higher in the tumor tissue than in resection margin (Fig. 7).

Bax and Bcl_2 mRNA were expressed both in tumor tissue and the resection margin. The Bcl_2 expression was stronger than that of Bax in the cancer tissue and the ratio of Bax to Bcl_2 was smaller in 10 of 12 tumors than at their resection margins. Therefore, the major difference in apoptosis-related gene expression between the cancer tissue and resection margin was enhanced level of mRNA for Bcl_2 rather than for Bax in cancer tissue when compared to resection margin.

DISCUSSION

This study confirms previous observations that colon tumors including low grade MALT lymphoma are strongly associated with CagA-positive HP infection [25,28–30] and shows that this infection in cancer patients is accompanied by an increased mRNA expression for gastrin and its receptors, for COX-2 and for anti-apoptotic Bcl_2 in the tumor tissues.

The reason for the higher HP seroprevalence in the colorectal cancer patients is not known but this infection results in a significant increase in plasma gastrin level possibly due to the impairment by HP of the local control of the G-cell activity by somatostatin in gastric antrum [31] or to direct stimulatory influence of the HP-originating histamine metabolite (Nαmethyl histamine), a potent luminally acting gastrin releaser [32,33]. It is of interest that increased rate of HP infection in colorectal cancer was estimated mainly from the positive serology anti-HP IgG, while UBT was positive in relatively smaller percentage of these patients (as compared to controls) probably because of the gastric mucosal atrophic changes that may interfere with the UBT as reported previously [21,34,35].

The question remains whether this higher HP seroprevalence plays any role in the pathogenesis of colorectal cancer but the fact that this infection is accompanied by an increased plasma level of gastrin reported previously [30,31,34] and observed also in this study, suggests that this hormone could contribute to the colorectal cancerogenesis by inducing higher mucosal cell proliferation of colon [19]. As described by Kinzler and Vogelstein [36] the colon cancer development starts with hyperplasia/dysplasia, progressing through adenoma to carcinoma (see Fig. 2). This finally leads to induction of COX-2 as it happens in other cancers [34–39]. Previous studies showed that gastrin gene is expressed by colorectal cell lines and by primary colon cancer [40–44] suggesting an autocrine role

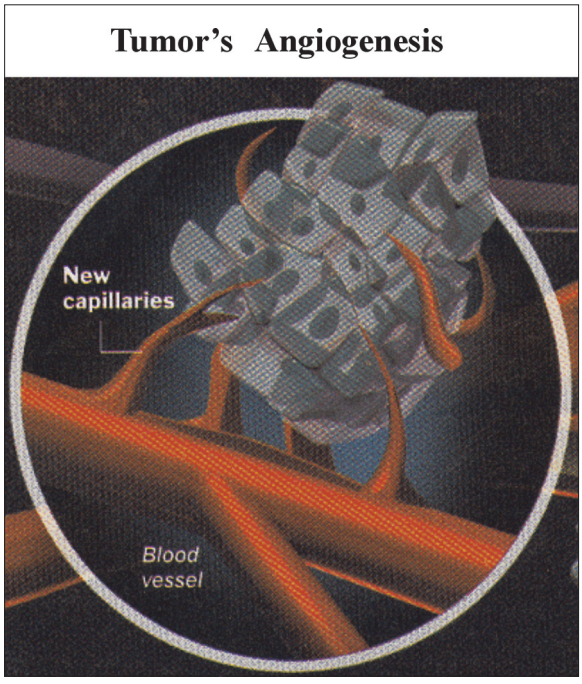

Figure 8. Angiogenesis is stimulated by COX-2 and its products including prostaglandins, VEGF, TGFα, EGF, bFGF, angiopoetins and other angiogenic substances involved in tumor growth and progression. This process depends upon the induction of COX-2 probably by progastrin and other growth factors generated in the cancer tissue. COX-2 overexpression results in the growth of endothelial cells and formation of new capillaries in the growing tumor
Blocking COX-2 by non-specific agents such as aspirin or by more specific COX-2 blockers (e.g. Vioxx lub Celecoxib), limits angiogenesis and slows down the tumor growth. Suppression of angiogenesis or the application of agents that diretcly reduce the amount of blood flowing to the cancer (e.g. Neovastat, Semaxanib) results in the reduction in the blood flow to the cancers cells which then become the targets of immunological system. (modified from L. Tweeten, Time 2001).

of gastrin in the growth of colon cancer cells but plasma levels were either not measured in those studies [25–28].

The main finding of the present study is the observation that the colorectal cancer tissue and resection margin express mRNA for gastrin and CCK_B-R and contain several fold higher content of immunoreactive amidated gastrin than the intact colonic mucosa. This indicates that the greatly increased gastrin in the plasma originated predominantly from the colorectal cancer and that besides the antral G-cells, the colorectal cancer tissue could contribute, to the increased plasma levels of this hormone though the fact that following removal of cancer there was relatively small decrease in plasma gastrin, was observed militates against the major involvement of gastrin-producing colorectal cancer cells to the elevated plasma hormone concentration in cancer patients.

Our finding that colorectal cancers exhibit higher expression and content of gastrin and its receptors reminds similar upregulation of gastrin biosynthesis described previously by our group in gastric cancers [21,39]. The major difference between gastric cancer and colorectal cancer is an overexpression of gastrin that seems to be the major product of the posttranslational changes of pre-progastrin in colorectal but not in gastric cancer tissue. As gastrin is known to be the powerful mucosal growth promoting factor in gastrointestinal tract [17,18,23] it is tempting to assume that this hormone could play a key role in the initiation and the progression of cancer disease both in the stomach and in the colon, both originating embryologically from the same endoderm tube [45]. This is supported by recent reports that gastrin peptides and their receptors are invariably coexpressed in the adenocarcinoma of the pancreas [46] or the liver [47] that also originates from the same endoderm as the stomach and gut. This study suggests that gastrin produced by pancreatic cancer cells may be an autocrine factor in carcinoma arising also from the pancreatic acinar cells. These studies are of particular interest because they show that in addition to amidated gastrin, glycine-extended gastrins as well as N-terminal progastrins were detected in the pancreatic and liver cancer and all appear to exhibit the growth promoting effects as amidated gastrin itself. Recently, Singh et al, [48] using transgenic mice overexpressing either progastrin or amidated gastrin found that the risk of colonic cancerogenesis is significantly increased in mice expressing progastrin rather than amidated gastrin. It is not clear whether in colorectal cancer in humans the increased levels of progastrin enhance the risk for developing of adenocarcinoma in the colon [23] but our results are in keeping with that noition.

Epidemiological studies indicate that the use of aspirin and other NSAID, whose major target is cyclooxygenase, a rate limiting enzyme converting arachidonic acid to prostanoids, decreases the incidence and mortality from colorectal cancers [11–16] (Fig. 8). COX-2 has been found to occur frequently in the colorectal cancer where it is held responsible for the production of excessive amounts of PG, tumor cell proliferation, angiogen-

esis, reduction in apoptosis and tumor invasiveness [15,16]. Assuming that HP and gastrin are initiators of colorectal carcinogenesis, the excessive production of prostanoids might be implicated in the promotion of this process and could be a target for the chemotherapy using specific COX-2 inhibitors [14–17]. Our results confirm the gene expression of both COX-1 and COX-2 in the colorectal cancer and thus, supports the notion that, indeed, COX-1 and COX-2 derived prostanoids may play a role in the development of colorectal cancer and should be a target of treatment using specific COX-2 inhibitors. As the elimination of the initiator of colorectal cancerogenesis such as increased progastrin and gastrin production may not be at present possible, the inhibition of the promotion of this process, particularly of angiogenesis and tumor growth could be attained by application of novel anti-inflammatory drugs. Based on our data that the HP is commonly infecting the colorectal cancer patients and leads to hypergastrinemia and COX-2 expression, both factors probably responsible for the initiation and promotion of colorectal cancer, it seems advisable to eradicate the HP in cancer patients to prevent or to attenuate the progression of colorectal cancer.

The growth rate of colorectal tumors, like other neoplasia, depends upon the perturbation in the balance between the cell division and cell death through apoptosis [49,50].

Several families of proteins have ability to modulate apoptosis, perhaps the most well known of which is the Bcl_2 that appears to repress apoptosis and Bax that promotes this process. Overexpression of Bcl_2 has been reported in a wide variety of cancers including colorectal, lung, prostate and other solid tumors and Bcl_2 has been held responsible for the blockade of the programmed cell death [51–53] and consequently for the tumor growth and relative resistance to chemotherapeutic drugs. Our study confirms previous reports [53,54] that the mRNA expression for Bcl_2 dominates in colorectal cancer tissues and shows for the first time that Bcl_2 mRNA is expressed more intensively in colorectal tumors, while Bax mRNA is expressed to similar extent in both the tumor and the resection margin of this tumor. As tumor tissue was also found to produce greater amounts of gastrin and to express more COX-2 than the resection margin, it may be reasonably to assume that the primary event in colorectal cancerogenesis is an excessive local biosynthesis of gastrin (progastrin and amidated gastrin) that in turn, as a potent mitogen, could induce COX-2 expression resulting in the reduction in apoptosis in the tumor tissue (Fig. 9). This requires normal activity of P53, which is considered as a guardian of genome. When P53 is mutated and inactivated, the damage of DNA is nor repaired and replication of such damaged DNA continues with tumor growth. The decrease in apoptosis in the cancer tissue might be responsible for the uncontrolled cell proliferation and tumor growth. The overexpression of COX-2 and related overproduction of PG and angiogenic factors that might be responsible for further stimulation of cancer growth, angiogenesis and invasiveness could represent a potential target of chemotherapy by COX-2 specific blockers but this requires adequate clinical trials.

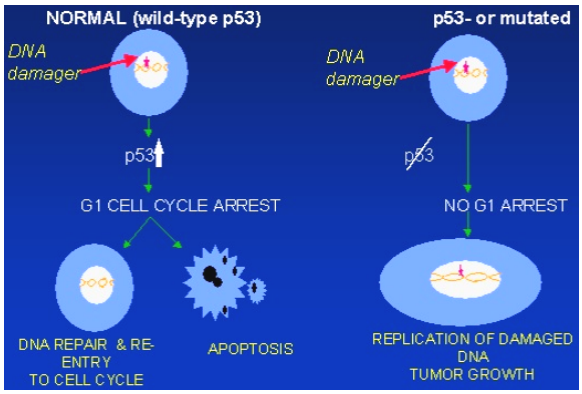

Figure 9. P53 - guardian of genom

CONCLUSIONS

1. Colorectal carcinoma and its resection margin overexpress gastrin and receptors for gastrin (CCK_B-R), and COX-2.

2. Here, we propose that an increased plasma level of gastrin should be considered as suitable biomarker of colorectal cancer;

3. HP infection may contribute to colonic cancerogenesis by enhancing expression of gastrin and COX-2, they may account for stimulation of the tumor growth, angiogenesis and reduction in apoptosis as evidenced by an increased ratio of mRNA expression for anti-apoptotic Bcl_2 over proapoptotic Bax proteins; and

4. HP positive patients who develop colorectal cancer should be subjected to the HP eradication; this is expected to reduce hypergastrinemia and to attenuate COX-2 expression. Our final conclusion would be: treatment of patients with

colorectal cancer with COX-2 selective inhibitors now gained a strong support as a preventive measure.

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